

In vitro* culture of *Gentiana kurroo

Agnieszka Fiuk, Jan J. Rybczyński

Botanical Garden – Centre for Biological Diversity Conservation, Polish Academy of Sciences, Warszawa

Callus cultures of *Gentiana kurroo* were initiated from hypocotyles, roots and cotyledons of 16-day old seedlings. Initiation agar medium was completed by MS salts supplemented with $1.0 \text{ mg}\cdot\text{l}^{-1}$ kinetin (KIN) and $0.5 \text{ mg}\cdot\text{l}^{-1}$ 2,4-dichlorophenoxyacetic acid (2,4-D). The most abundant callus proliferation was observed on root and hypocotyle explants, after 30 days of culture. Cotyledon proliferation was connected only with surfaces of the cutting. Besides callus proliferation, somatic embryo formation was observed, too. For cell suspension initiation and maintaining, MS medium supplemented with $2.0 \text{ mg}\cdot\text{l}^{-1}$ 6-benzylaminopurine (BAP), $1.0 \text{ mg}\cdot\text{l}^{-1}$ Dicamba and $0.1 \text{ mg}\cdot\text{l}^{-1}$ 1-naphthaleneacetic acid (NAA) was used. Cell suspension growth curves were done. The best results were obtained in cotyledon-derived suspension.

To describe the morphogenetic potential of maintained suspensions, a portion of 100 mg of fractionated tissue was implanted on MS agar medium supplemented with $80 \text{ mg}\cdot\text{l}^{-1}$ adenine sulphate and various concentrations of KIN and GA_3 . The highest and the lowest number of somatic embryos were regenerated by the cotyledon and root-derived suspension, respectively. The size of aggregates, and KIN and GA_3 combinations of concentrations used to had significant influence on somatic embryo production.

The suspensions of the highest regeneration potential has been selected for protoplast isolation, culture and plant regeneration experiments. Three methods of protoplast culture were employed: liquid, agarose solidified and bead culture with three different media. Two ways of protoplast development were observed: protocallus formation and somatic embryo regeneration. The DNA of somatic embryo derived regenerants was described with the help of flow cytometry.

X Ogólnopolska Konferencja Kultur In Vitro i Biotechnologii Roślin
15 - 17 września 2003 r Bydgoszcz
Biotechnologia Roślinna w Biologii, Farmacji i Rolnictwie